

## Microbial Pectinolytic Enzymes

Ghellai Lotfi\*

Laboratory of Food and Environmental Microbiology (LAMAABE), University of Tlemcen, 13000 Tlemcen, Algeria,  
Department of Biology, University of Saïda, 20100 Saïda, Algeria

Received: 31 April 2016; Accepted: 02 June 2016

---

### Abstract

Apart from plant, microorganisms such as bacteria and fungi are currently well known to be able to hydrolyze pectic substances like pectin, protopectin and pectic acids, thanks to a various forms of pectinolytic enzymes. These enzymes have been intensively used in various fields. The present review is focused mainly on microbial pectinolytic enzymes.

**Keywords:** pectic substances, pectinases, bacteria, fungi, yeast.

---

### 1. Introduction

Pectic substances and celluloses are the most abundant carbohydrates present in plants (Table 1). Pectic substances, present in cell wall and middle lamella, contribute firmness and structure to plant tissues [1]. According to the American Chemical Society pectic substances are classified into four main types [2]: (I) Protopectin, (II) Pectic acid, (III) Pectinic acids, (IV) Pectin (Polymethyl galacturonate). Pectinases or pectinolytic enzymes, a heterogeneous group of related enzymes, are responsible for hydrolyze of pectic substances present in plant tissues. Pectinases are one of the most widely distributed enzymes in microorganisms (bacteria and fungi) and higher plants [3]. Indeed, It has been shown in the literature that pectinases account for 25% of the global food enzymes sales.

Almost all the commercial preparations of pectinases are produced from fungal sources [4]. *Aspergillus niger* is the most commonly used fungal species for industrial production of pectinolytic enzymes [5, 6].

Pectinolytic enzymes are of significant importance in the current biotechnological era with their all embracing applications in fruit juice extraction and its clarification, scouring of cotton, degumming of plant fibers, waste water treatment, vegetable oil extraction, tea and coffee fermentations, bleaching of paper, in poultry feed additives and in the alcoholic beverages and food industries [7]. Protopectinases, polygalacturonases, lyases and pectin esterases are among the extensively studied pectinolytic enzymes.

### 2. Occurrence of pectinases

#### 2.1. Protopectinases

These enzymes catalyze the solubilization of protopectin. They are found in the culture filtrates of yeast and yeast-like fungi. They have been isolated from *Kluyveromyces fragilis* IFO 0288, *Galactomyces reesei* L., *Trichosporon penicillatum* SNO 3 [3] and from the culture filtrate of a wide range of *Bacillus* sp. [8]. Protopectinases have also been found in *Trametes* sp. [9].

Table 1. Percentages of pectic substances in fruits and vegetables [7]

Fruit/vegetable	tissue	Pectic substance (%)
Apple	Fresh	0.5–1.6
Banana	Fresh	0.7–1.2
Peaches	Fresh	0.1–0.9
Strawberries	Fresh	0.6–0.7
Cherries	Fresh	0.2–0.5
Peas	Fresh	0.9–1.4
Carrots	Dry matter	6.9–18.6
Orange pulp	Dry matter	12.4–28.0
Potatoes	Dry matter	1.8–3.3
Tomatoes	Dry matter	2.4–4.6
Sugar beet pulp	Dry matter	10.0–30.0

## 2.2. Polygalacturonases

Polygalacturonases hydrolyze the polygalacturonic acid chain by addition of water and are the most abundant among all the pectinolytic enzymes. Endo-polygalacturonases are widely distributed among fungi, bacteria and many yeasts [10]. They have been reported in many microorganisms, including *Aureobasidium pullulans* [11], *Rhizoctonia solani* Kuhn [12], *Fusarium moniliforme* [13], *Neurospora crassa* [14], *Rhizopus stolonifer* [15], *Aspergillus* sp. [16], *Thermomyces lanuginosus* [17], *Peecilomyces clavisporus* [18]. Endo-polygalacturonases are also found in higher plants and some plant parasitic nematodes [9].

Exo-polygalacturonases have been reported in *Erwinia carotovora* [19], *Agrobacterium tumefaciens* [20], *Bacteroides thetaiotamicron* [21], *Erwinia chrysanthemi* [22], *Alternaria mali* [23], *Fusarium oxysporum* [24], *Ralstonia solanacearum* [25], *Bacillus* sp. [22].

## 2.3. Lyases

Lyases catalyze the trans-eliminative cleavage of the galacturonic acid polymer. Polygalacturonate lyases (or pectate lyases) are produced by many bacteria and some pathogenic fungi. Lyases have been isolated from bacteria and fungi associated with food spoilage and soft rot. They have been reported in *Colletotrichum lindemuthionum* [26], *Bacteroides thetaiotaomicron* [27], *Erwinia carotovora* [28], *Amucala* sp. [29], *Pseudomonas syringae* pv. *glycinea* [30], *Colletotrichum magna*

[31], *Erwinia chrysanthemi* [32, 33], *Bacillus* sp. [34, 35], *Bacillus* sp. DT-7 [36], *Colletotrichum gloeosporioides* [37, 38]. They have been reported to be produced by *Aspergillus japonicus* [39], *Penicillium paxilli* [40, 41], *Penicillium* sp. [42,43,44], *Pythium splendens* [45] *Pichia pinus* [46], *Aspergillus* sp. [47], *Thermoascus aurantiacus* [48].

## 2.4. Pectinesterases

This enzyme liberates pectins and methanol by de-esterifying the methyl ester linkages of the pectin backbone. Pectinesterase are found in plants, plant pathogenic bacteria and fungi [49]. It has been reported in *Rhodotorula* sp. [50], *Phytophthora infestans* [51], *Erwinia chrysanthemi* B341 [52], *Saccharomyces cerevisiae* [53], *Lachnospira pectinoschiza* [54], *Pseudomonas solanacearum* [55], *Aspergillus niger* [56,57], *Lactobacillus lactis* subsp. *Cremoris* [58], *Penicillium frequentans* [59], *Erwinia chrysanthemi* 3604 [60], *Penicillium occitanis* [61], *Aspergillus japonicus* [62] and others. There are many reports of occurrence of pectinesterase in plants.

## 3. Conclusion

The probable reason for the existence of a great diversity of pectinolytic organisms is that the pectic substances could be present in various forms in the nature. To date, certain species of bacteria, molds, and yeast are commonly studied for their aptitude to produce pectinases among others. However, *Aspergillus niger* remains, currently the most used fungal specie for industrial production of pectinolytic

enzymes. Therefore, the discovery of other microorganisms having a stable ability to produce such enzymes would constitute an enormous progress in biotechnology.

**Compliance with Ethics Requirements.** Authors declare that they respect the journal's ethics requirements. Authors declare that they have no conflict of interest and all procedures involving human / or animal subjects (if exist) respect the specific regulation and standards.

## References

1. Gummadi, S.N.; Panda, T., Purification and biochemical properties of microbial pectinases: a Review, *Process Biochemistry* **2003**, *38*, 987-996.
2. Alkorta, I.; Garbisu, C., Llama, M.J., Serra J.L., Industrial applications of pectic enzymes: a Review, *Process Biochem* **1998**, *33*, 21–8.
3. Whitaker, J.R., Microbial pectinolytic enzymes. In: Fogarty WM, Kelly CT, editors. Microbial enzymes and biotechnology. 2nd ed. London: Elsevier Science Ltd.; **1990**. p. 133–76.
4. Singh, S.A.; Ramakrishna, M., Rao, A.G.A., Optimization of downstream processing parameters for the recovery of pectinase from the fermented broth of *Aspergillus carbonarius*, *Process Biochem* **1999**, *35*, 411–7.
5. Kotzekidov, P., Production of polygalacturonases by *Byssachlamys fulva*, *J Ind Microbiol* **1991**, *7*, 53–6.
6. Naidu, G.S.N.; Panda, T., Production of pectolytic enzymes—a review, *Bioprocess Eng* **1998**, *19*, 355–61.
7. Jayani R.S.; Saxena, S., Gupta, R., Microbial pectinolytic enzymes: A review, *Process Biochemistry* **2005**, *40*, 2931–2944.
8. Sakai, T., Degradation of pectins. In: Winkelmann G, editor. Microbial degradation of natural products. Weinheim: VCH, **1992**. pp. 57–81.
9. Sakai, T.; Sakamoto, T., Hallaert, J., Vandamme, E.J., Pectin, pectinase and protopectinase: production, properties and applications, *Adv Appl Microbiol* **1993**, *39*, 231–94.
10. Luh, B.S.; Phaff, H.J., Studies on polygalacturonase of certain yeasts, *Arch Biochem Biophys* **1951**, *33*, 212–27.
11. Sakai, T.; Takaoka, A., Purification, crystallization and some properties of endo polygalacturonase from *Aureobasidium pullulans*, *Agric Biol Chem* **1984**, *49*, 449–58.
12. Marcus, L.; Barash, I., Sneh, B., Koltin, Y., Finkler, A., Purification and characterization of pectolytic enzymes produced by virulent and hypovirulent isolates of *Rhizoctonia solani* Kuhn, *Physiol Mol Plant Pathol* **1986**, *29*, 325–36.
13. De Lorenzo, G.; Salvi, G., Degra, L., D'Ovidio, R., Cervone, F., Introduction of extracellular polygalacturonases and its mRNA in the phytopathogenic fungus *Fusarium moniliforme*, *J Gen Microbiol* **1987**, *133*, 3365–73.
14. Polizeli, M.L.T.M.; Jorge, J.A., Terenzi, H.F., Pectinase production by *Neurospora crassa*: purification and biochemical characterization of extracellular polygalacturonase activity, *J Gen Microbiol* **1991**, *137*, 1815–23.
15. Manachini, M.; Fortina, C., Parini C., Purification and properties of an endopolygalacturonase produced by *Rhizopus stolonifer*, *Biotechnol Lett* **1987**, *9*, 219–24.
16. Nagai, M.; Katsuragi, T., Terashita, T., Yoshikawa, K., Sakai, T., Purification and characterization of an endo-polygalacturonase from *Aspergillus awamori*, *Biosci Biotechnol Biochem* **2000**, *64*, 1729–32.
17. Kumar, S.S.; Palanivelu, P., Purification and characterization of an exopolygalacturonase from the thermophilic fungus, *Thermomyces lanuginosus*, *World J Microbiol Biotechnol* **1999**, *15*, 643–6.
18. Souza, J.V.B.; Silva, T.M., Maia, M.L.S., Teixeira, M.F.S., Screening of fungal strains for pectinolytic activity: endopolygalacturonase production by *Peecilomyces clavispurus* 2A.UMIDA.1, *Process Biochem* **2003**, *4*, 455–8.
19. Palomäki, T.; Saarihahti, H.T., Isolation and characterization of new C-terminal substitution mutation affecting secretion of polygalacturonases in *Erwinia carotovora* ssp. *carotovora*, *FEBS Lett* **1997**, *400*, 122–6.
20. Rodrigues-Palenzuela, P.; Burr, T.J., Collmer, A., Polygalacturonase is a virulence factor in *Agrobacterium tumefaciens* biovar 3, *J Bacteriol* **1991**, *173*, 6547–52.
21. Tierny, Y.; Bechet, M., Joncquiert, J.C., Dubourguier, H.C., Guillaume, J.B., Molecular cloning and expression in *Escherichia coli* of genes encoding pectate lyase and pectin methylesterase activities from *Bacteroides thetaiotaomicron*, *J Microbiol* **1994**, *76*, 592–602.
22. Koboyashi, T.; Higaki, N., Yajima, N., Suzumatsu, A., Haghahara, H., Kawai, S., et al. Purification and properties of a galacturonic acidreleasing exopolygalacturonase from a strain of *Bacillus*, *Biosci Biotechnol Biochem* **2001**, *65*, 842–7.
23. Nozaki, K.; Miyairi, K., Hizumi, S., Fukui, Y., Okuno, T., Novel exopolygalacturonases produced by *Alternaria mali*, *Biosci Biotechnol Biochem* **1997**, *61*, 75–80.

24. Maceira, F.I.G.; Pietro, A.D., Roncero, M.I.G., Purification and characterization of a novel exopolygalacturonase from *Fusarium oxysporum* f. sp. *lycopersici*. *FEMS Microbiol Lett* **1997**, *154*, 37–43.
25. Huang, Q.; Allen, C., An exo-poly-a-D-galacturonosidase, Peh B, is required for wild type virulence of *Ralstonia solanacearum*, *J Bacteriol* **1997**, *179*, 7369–78.
26. Wijesundera, R.L.C.; Bailey, J.A., Byrde, R.J.W., Production of pectin lyase by *Colletotrichum lindemuthionum* in culture and in infected bean (*Phaseolus vulgaris*) tissue, *J Gen Microbiol* **1984**, *130*, 285–90.
27. McCarthy, R.E.; Kotarski, S.F., Salyers, A.A., Location and characteristics of the enzymes involved in the breakdown of polygalacturonic acid by *Bacteroides thetaiotaomicron*, *J Bacteriol* **1985**, *161*, 493–9.
28. Kotoujansky, A., Molecular genetics of pathogenesis by soft-rot *Erwinia*, *Annu Re Phytopathol* **1987**, *25*, 405–30.
29. Bruhlmann, F., Production and characterization of an extracellular pectate lyase from an *Amycolata* sp., *Appl Environ Microbiol* **1995**, *61*, 3580–5.
30. Margo, P.; Varvaro, L., Chilosì, G., Avanzo, C., Balestra, G.M., Pectinolytic enzymes produced by *Pseudomonas syringae* pv. *Glycinea*, *FEMS Microbiol Lett* **1994**, *117*, 1–6.
31. Wattad, C.; Freeman, S., Dinoor, A., Prusky, D., A nonpathogenic mutant of *Colletotrichum magna* is deficient in extracellular secretion of pectate lyase, *Molec Plant-Microbe Interac* **1995**, *8*, 621–6.
32. Favey, S.; Bourson, C., Bertheou, Y., Kotoujansky, A., Boccara, M., Purification of the acidic pectate lyase of *Erwinia chrysanthemi* 3937 and sequence analysis of the corresponding gene, *J Gen Microbiol* **1992**, *138*, 499–508.
33. Shevchik, V.E.; Robert-Baudouy, J., Hugouvieux-Cotte-Pattat, N., Pectate lyase Pel 1 of *Erwinia chrysanthemi* 3937 belongs to a new family, *J Bacteriol* **1997**, *179*, 7321–30.
34. Koboyashi, T.; Koike, K., Yoshimatsu, T., Higaki, N., Suzumatsu, A., Ozawa, T., et al. Purification and properties of a low-molecular weight, high-alkaline pectate lyase from an alkaliphilic strain of *Bacillus*, *Biosci Biotechnol Biochem* **1999**, *63*, 72–5.
35. Takao, M.; Nakaniwa, T., Yoshikawa, K., Terashita, T., Sakai, T., Molecular cloning, DNA sequence, and expression of the gene encoding for thermostable pectate lyase of thermophilic *Bacillus* sp. TS 47, *Biosci Biotechnol Biochem* **2001**, *65*, 322–9.
36. Kashyap, D.R.; Chandra, S., Kaul, A., Tewari, R., Production, purification and characterization of pectinase from a *Bacillus* sp. DT7, *World J Microbiol Biotechnol* **2000**, *16*, 277–82.
37. Yakoby, N.; Kobiler, I., Dinoor, A., Prusky, D., pH regulation of pectate lyase secretion modulates the attack of *Colletotrichum gloeosporioides* in avocado fruits, *Appl Environ Microbiol* **2000**, *66*, 1026–30.
38. Drori, N.; Kramer-Haimovich, H., Rollins, J., Dinoor, A., Okon, Y., Pines, O., et al. External pH and nitrogen source affect secretion of pectate lyase by *Colletotrichum gloeosporioides*, *Appl Environ Microbiol* **2003**, *69*, 3258–62.
39. Ishii, S.; Yokotsuka, T., Purification and properties of pectin lyase from *Aspergillus japonicus*, *Agric Biol Chem* **1975**, *39*, 313–21.
40. Szajer, I.; Szajer, C., Formation and release of pectin lyase during growth of *Penicillium paxilli*, *Biotechnol Lett* **1985**, *7*, 105–8.
41. Szajer, I.; Szajer, C., Pectin lyase of *Penicillium paxilli*, *Biotechnol Lett* **1982**, *4*, 549–52.
42. Diaz-Borras, M.A.; Aguilar, R.V., Glimenez, E.H., Comparación de la actividad pectinolítica in vitro entre cepas de *Penicillium* sensibles y resistentes a fungicidas comerciales, *Alimentaria* **1987**, *24*, 57–8.
43. Alana, A.; Alkorta, I., Dominguez, J.B., Llama, M.J., Serra, J.L., Pectin lyase activity in a *Penicillium italicum* strain, *Appl Environ Microbiol* **1990**, *56*, 3755–9.
44. Sapunova, L.I.; Mikhailova, R.V., Lobanok, A.G., Properties of pectin lyase preparations from the genus *Penicillium*, *Appl Microbiol Biochem* **1995**, *31*, 435–8.
45. Chen, W.C.; Hsieh, H.J., Tseng, T.C., Purification and characterization of a pectin lyase from *Pythium splendens* infected cucumber fruits, *Botanical Bull Academia Sinica* **1998**, *39*, 181–6.
46. Moharib, S.A.; El-Sayed, S.T., Jwanny, E.W., Evaluation of enzymes produced from yeast, *Nahrung* **2000**, *44*, 47–51.
47. Sunnotel, O.; Nigam, P., Pectinolytic activity of bacteria isolated from soil and two fungal strains during submerged fermentation, *World J Microbiol Biotechnol* **2002**, *18*, 835–9.
48. Martins, E.S.; Silva, D., Da Silva, R., Gomes, E., Solid state production of thermostable pectinases from thermophilic *Thermoascus aurantiacus*, *Process Biochem* **2002**, *37*, 949–54.
49. Hasunuma, T.; Fukusaki, E.I., Kobayashi, A., Methanol production is enhanced by expression of an *Aspergillus niger* pectin methylesterase in tobacco cells, *J Biotechnol* **2003**, *106*, 45–52.

50. Vaughan, I.L.H.; Jakubczly, T., McMillan, J.D., Higgins, T.E., Dave, B.A., Crampton VM. Some pink yeasts associated with softening of olives, *Appl Microbiol* **1969**, *18*, 771–5.
51. Forster, H., Pectinesterase from *Phytophthora infestans*, *Methods Enzymol* **1988**, *161*, 355–7.
52. Pitkänen, K.; Heikinheimo, R., Pakkanen, R., Purification and characterization of *Erwinia chrysanthemi* B374 pectin methylesterase produced by *Bacillus subtilis*, *Enzyme Microbial Technol* **1992**, *14*, 832–6.
53. Gainvors, A.; Frezier, V., Lemaesquier, H., Lequart, C., Aigle, M., Belarbi, A., Detection of polygalacturonase, pectin lyase and pectinesterase activities in *Saccharomyces cerevisiae* strain, *Yeast* **1994**, *10*, 1311–9.
54. Cornick, N.A.; Jensen, N.S., Stahl, D.A., Hartman, P.A., Allison, M.J., *Lachnospira pectinoschiza* sp. nov., an anaerobic pectinophile from the pig intestine, *Int J Syst Bacteriol* **1994**, *44*, 87–93.
55. Schell, M.A.; Denny, T.P., Huang, J., Extracellular virulence factors of *Pseudomonas solanacearum*: role in disease and their regulation. In: Kado CI, Crosa JH, editors. *Mol Mech Bacterial Virulence*. The Netherlands: Kluwer Academic Press; **1994**, pp. 311–24.
56. Maldonado, M.C.; Saad, A.M.S., Callieri, D.A.S., Purification and characterization of pectinesterase produced by a strain of *Aspergillus niger*, *Curr Microbiol* **1994**, *24*, 193–6.
57. Maldonado, M.S., Saad, A.M.S., Production of pectinesterase and polygalacturonase by *Aspergillus niger* in submerged and solid state systems, *J Ind Microbiol Biotechnol* **1998**, *20*, 34–8.
58. Karam, N.E.; Belarbi, A., Detection of polygalacturonase and pectinesterases in lactic acid bacteria, *World J Microbiol Biotechnol* **1995**, *11*, 559–63.
59. Kawano, C.Y.; Chellegatti, M.A.S.C., Said, S., Fonseca, M.J.V., Comparative study of intracellular and extracellular pectinases produced by *Penicillium frequentans*, *Biotechnol Appl Biochem* **1999**, *29*, 133–40.
60. Laurent, F.; Kotoujansky, A., Bertheau, Y., Overproduction in *Escherichia coli* of the pectin methylesterase A from *Erwinia chrysanthemi* 3937: one-step purification, biochemical characterization, and production of polyclonal antibodies, *Can J Microbiol* **2000**, *46*, 474–80.
61. Hadj-Taieb, N.; Ayadi, M., Trigui, S., Bouabdollah, F., Gargouri, A., Hyper production of pectinase activities by fully constitutive mutant (CT 1) of *Penicillium occitanis*, *Enzyme Microbial Technol* **2002**, *30*, 662–6.
62. Semenova, M.V.; Grishutin, S.G., Gusakov, A.V., Okunev, O.N., Sinitsyu, A.P., Isolation and properties of pectinases from the fungus *Aspergillus japonicus*, *Biochemistry* **2003**, *68*, 559–69.